

ON THE UTILIZATION OF XYLOSE BY THE WHITE RAT

BY
MABEL MARIE MILLER

A DISSERTATION SUBMITTED IN PARTIAL FULFILMENT OF THE
REQUIREMENTS FOR THE DEGREE OF DOCTOR OF PHI-
LOSOPHY IN THE UNIVERSITY OF MICHIGAN

1932

REPRINTED FROM

THE JOURNAL OF BIOLOGICAL CHEMISTRY
VOL. XCVIII, No. 1, PAGES 133-150, OCTOBER, 1932

ON THE UTILIZATION OF XYLOSE BY THE WHITE RAT

BY
MABEL MARIE MILLER

A DISSERTATION SUBMITTED IN PARTIAL FULFILMENT OF THE
REQUIREMENTS FOR THE DEGREE OF DOCTOR OF PHI-
LOSOPHY IN THE UNIVERSITY OF MICHIGAN

1932

REPRINTED FROM

THE JOURNAL OF BIOLOGICAL CHEMISTRY
VOL. XCVIII, No. 1, PAGES 133-150, OCTOBER, 1932

*27699



PENTOSE METABOLISM

I. THE RATE OF ABSORPTION OF *d*-XYLOSE AND THE FORMATION OF GLYCOGEN IN THE ORGANISM OF THE WHITE RAT AFTER ORAL ADMINISTRATION OF *d*-XYLOSE*

By MABEL M. MILLER AND HOWARD B. LEWIS

(From the Department of Physiological Chemistry, Medical School, University of Michigan, Ann Arbor)

(Received for publication, July 8, 1932)

Xylose, first discovered and isolated from wood by Koch in 1881 (1), has been one of the rarer sugars, since it could be prepared only with difficulty and at considerable cost. In 1930, however, the United States Bureau of Standards in a cooperative investigation announced a simple method of preparation of xylose (2) from cottonseed hull bran, a very cheap raw material. Xylose thus becomes a sugar obtainable at a price comparable to that of sucrose. The wide publicity given to this announcement of the cheap production of xylose on a semicommercial scale has led to statements concerning xylose in the current popular literature which may be misleading. Thus xylose has been characterized (3) as a "slenderizing sugar from cottonseed" and, in the same article, as a "non-fattening sugar." Since in popular usage the term sugar is usually considered to imply a substance utilizable as a food by the higher forms of animal life, it becomes of importance to raise again the question of the rôle of xylose (and the pentoses) in nutrition. Early in the progress of the work, through the courtesy of Professor James L. Kassner of the University of Alabama and of the Bureau of Standards, a supply of this new product was placed at our disposal for a study of this problem.

Many conflicting conclusions have been drawn by various workers concerning the extent and mechanism of the utilization of the pentoses. The work has been limited hitherto by a lack of uni-

* A preliminary report of a part of this investigation appears in *Proc. Soc. Exp. Biol. and Med.*, **28**, 448 (1930-31).

formity in the experimental procedure, by inadequate analytical methods, and by the cost of the pentoses to be studied. Pflüger (4) in 1905 reviewed and severely criticized the older work which indicated that pentoses could give rise to glycogen in the animal body and concluded that no adequate experimental evidence was available in support of this belief. Of later workers in this field, Brasch (5) observed no glycogen formation in dogs or rabbits after oral administration of rhamnose, arabinose, or xylose. On the other hand, Thomas, Gradinescu, and Imas (6) in experiments with frogs and Nitzescu and Benetato (7) in experiments with rabbits observed an increased glycogen storage after the administration of the five carbon sugars. In agreement with older work, the most effective source of glycogen derived from pentoses was rhamnose.

This brief review serves to illustrate the contradictory nature of the evidence concerning the transformation of the pentoses into glycogen. In these experiments, the species of experimental animals were frequently different, the amounts of pentose fed varied, the preliminary treatment of the animals was not uniform, and no knowledge as to whether the pentose fed was actually absorbed from the intestine was available. It was impossible to compare absorption of and glycogen formation from the pentoses with those of a readily utilizable sugar such as glucose.

By the new method of study introduced by Cori (8), it has become possible to determine with considerable accuracy not only the extent of absorption but also the rate at which absorption has occurred. With such additional data available, studies of glycogen formation assume a greater significance. In the present paper the rate of absorption of xylose from the intestinal tract of the white rat has been studied by the procedure of Cori (8) and in the same animals, the extent of glycogen deposition has been determined. Further studies of the fate of the xylose absorbed will be presented in a subsequent paper.

EXPERIMENTAL

Young white rats weighing from 100 to 200 gm. were fasted for 24 hours. A known amount of xylose (determined by the technique employed by Cori) was introduced by stomach tube and after the desired interval of time, the animals were killed with chloroform. The entire gastrointestinal tract was exposed, ligated

at each end, and removed. The tract was split open and the contents were washed out with several portions of hot water into a volumetric flask of 100 cc. capacity. After cooling, 5 cc. of 10 per cent sodium tungstate and a like amount of 0.66 N sulfuric acid were added, the contents of the flask were diluted to the mark with water, and the precipitated proteins were filtered off. The filtrates were used for the determination of the amount of unabsorbed pentose.

Xylose was determined by the method of Hagedorn and Jensen. Since the values obtained in this method are given in terms of glucose, it was necessary to determine the reduction effected by known amounts of xylose. After these values had been obtained and plotted, a table of reduction values in terms of xylose was constructed from the graph and these figures were used in subsequent determinations.

One group of seventeen rats served as controls for determining the normal reduction values of the contents of the gastrointestinal tract after a 24 hour fast. Values ranging from 8.3 to 16.3 mg. with an average value of 11.2 mg. (calculated as xylose) were obtained. In the xylose feeding experiments, all values for residual sugar in the gastrointestinal tract were accordingly corrected for the normal fasting reduction value equivalent to 11 mg. of xylose.

In a series of check experiments, the accuracy of the experimental procedure was tested as follows: Rats were fed xylose in the usual manner (approximately 2 cc. of a 50 per cent solution of xylose), killed immediately without allowing any time interval for the absorption of the carbohydrate, and the contents of the tract were analyzed. In ten experiments in which amounts of xylose varying from 700 to 1030 mg. were fed, the absolute error in the amount of xylose recovered varied from +19 to -21 mg., with an average percentage recovery of 99.5 per cent of the xylose fed.

Most of the rats used for the determination of the absorption coefficient were used also for glycogen estimations. Therefore, after the removal of the gastrointestinal tract from 5 to 10 minutes were needed for the preparation of the tissues for glycogen analysis. In some cases the analysis of the gastrointestinal contents was begun within 10 to 15 minutes after the death of the rat, but in most instances, the gastrointestinal tracts as removed from the animals were placed in covered beakers and kept in the ice box

TABLE I

Rate of Absorption of Xylose and Glucose and Formation of Glycogen after Oral Administration of Xylose and Glucose

Sugar administered	Rat No.	Weight after 24 hr. fast	Absorption period	Sugar fed	Sugar absorbed per 100 gm. rat per hr.	Glycogen	
		gm.	hrs.			Liver	Body
	Average of 11 rats (controls)	103		0	0	0.11 (0.07-0.17) *	0.05 (0.04-0.08)
Xylose	Average of 6 rats	169	1	520-930	29 (25-34)	0.16 (0.09-0.27)	0.05 (0.05-0.06)
"	" "	149	2	530-1010	39 (29-54)	0.13 (0.09-0.19)	0.06 (0.04-0.08)
Xylose	19	94	3	570	74	0.13	0.05
	20	107	3	570	50	0.15	0.07
	21	106	3	570	56	0.16	0.08
	23	108	3	390	34	0.19	0.07
	25	111	3	390	33	0.16	0.06
	26	123	3	390	33	0.14	0.07
	27	129	3	560	49	0.11	0.04
	28	116	3	560	43	0.15	0.06
	29	137	3	560	41	0.17	0.05
	30	122	3	560	38	0.15	0.05
	31	103	3	560	52	0.08	0.07
	32	118	3	560	54	0.06	0.04
Average.....		114			46	0.14	0.06
Glucose	47	118	3	725	200	2.00	0.15
	50	96	3	725	231	2.00	0.19
	95	154	3	800	144	1.70	0.10
	96	151	3	800	147	1.54	0.10
	97	147	3	800	151	1.37	0.11
	98	159	3	800	149	1.18	0.10
	103	150	3	890	165	3.08	0.26
	104	158	3	890	145	2.91	0.24
Average.....		141			166	1.97	0.16

* The figures in parentheses indicate the maximal and minimal values observed in each series.

TABLE I—*Concluded*

Sugar administered	Rat No.	Weight after 24 hr. fast	Absorption period	Sugar fed	Sugar absorbed per 100 gm. rat per hr.	Glycogen	
						Liver	Body
		<i>gm.</i>	<i>hrs.</i>	<i>mg.</i>	<i>mg.</i>	<i>per cent</i>	<i>per cent</i>
Glucose†	145	130	3	177	44	0.54	0.09
	146	139	3	177	42	0.56	0.08
	147	131	3	177	44	0.63	0.09
	149	168	3	226	44	0.57	0.08
	150	174	3	226	42	0.57	0.08
	151	175	3	226	42	0.59	0.08
	154	169	3	266	52	0.69	0.09
	155	163	3	266	54	0.64	0.10
	156	166	3	266	52	0.75	0.10
	157	179	3	266	49	0.72	0.10
	158	174	3	266	50	0.66	0.10
Average.....		160			47	0.63	0.09

† The animals in this series received glucose in such an amount that the absorption in 3 hours is comparable to the absorption of xylose during the same time interval.

until all the rats for that day had been killed. When this procedure was followed, analysis of the gastrointestinal contents was begun within 1.5 to 2 hours after the death of the rat. No noticeable differences in the xylose content within the same groups were found due to the longer time interval before analysis was begun. Therefore, it can be assumed that bacterial action, if any took place, was too small to be significant.

As in previous experiments from this laboratory (9), in the glycogen determinations (Pflüger), the glucose formed by the hydrolysis of the glycogen was estimated by the method of Hagedorn and Jensen and the factor of Pflüger (0.927) was used for the conversion of glucose into glycogen. In Table I, under the heading "body glycogen" is given the glycogen content of the entire organism minus the liver and gastrointestinal canal.

The xylose as received from the laboratories of the University of Alabama and the Bureau of Standards was slightly yellow in color. This product was recrystallized twice from hot ethyl alcohol to which a small amount of norit was added. The final white product

was dried in a vacuum oven at 40° and then in a desiccator over calcium chloride.

DISCUSSION

The experimental groups consisted of six series of animals, a fasting control group of eleven rats, three groups which were killed 1, 2, and 3 hours respectively after the administration of the xylose, and two control groups which received glucose and were killed after 3 hours. Since the results of the 3 hour absorption periods were considered most significant, the figures for these groups alone are presented in detail in Table I, the results with the other groups being summarized.

The absorption coefficients of xylose showed a tendency to increase as the absorption period was prolonged. Average values of 29, 39, and 46 mg. of xylose absorbed per hour per 100 gm. of rat were observed during absorption periods of 1, 2, and 3 hours respectively. We found it impractical to continue absorption studies beyond 3 hours because of the frequent development of diarrhea. Cori (8) obtained an absorption coefficient of 62 mg. in the 1st hour and a lower but nearly uniform rate of absorption subsequently, 24, 28, 32, and 28 mg. at the end of absorption periods of 2, 3, 4, and 5 hours duration respectively. In none of our group of six animals did the rate of absorption in the 1st hour approach that found for the same period by Cori. Macleod, Magee, and Purves (10) were able to show that when concentrated solutions of glucose were fed, they were diluted in the stomach by the gastric secretion to a concentration of approximately 0.75 M before being discharged into the intestine and that the rate of discharge of such sugar solutions through the pylorus was inversely proportional to their concentration. If these findings are applicable also to xylose, a slower absorption of xylose during the 1st hour after feeding might have been expected in both Cori's and our own experiments, since the solutions of xylose fed were much more concentrated than 0.75 M (approximately 1.5 to 3.0 M in our series) and some delay in discharge through the pylorus would presumably result because of the period required for dilution. Our results, in general, lend support to the findings of Macleod and his coworkers. We have also observed that our

animals, which were allowed free access to water at all times, drank frequently after the feeding of the concentrated solutions of the sugars.

Inspection of the data concerning the glycogen content of the liver and the rest of the body, fails to reveal any appreciable increase in glycogen content after xylose feeding as compared with the fasting controls. On the other hand, when glucose was fed in amounts similar to xylose, the deposition of glycogen was marked both in the liver and in the rest of the body. However, since the rate of absorption of glucose was so much higher than that of xylose, it was considered necessary as a final control to investigate the glycogenetic value of glucose, when this sugar was fed at such a level that the amount absorbed in 3 hours would approximate the amount of xylose absorbed in the same period. In such experiments the amounts of carbohydrate available for glycogenesis would be approximately the same for both glucose and xylose. In this series (Table I) the average rate of absorption per 100 gm. of rat per hour was 47 mg. of glucose, a figure comparable to the average value of 46 mg. in the 3 hour absorption experiments with xylose. From a comparison of the glycogen values with those of the control and of the 3 hour xylose absorption periods, it is evident that a very considerable formation of glycogen has occurred even with smaller amounts of glucose.

When such smaller amounts of glucose were fed, a large part of the glucose was undoubtedly absorbed very promptly. It was felt that the greater glycogen formation from glucose fed at the level of xylose absorption, as compared with the xylose experiments, might have been due to this more rapid absorption of glucose with the resulting longer period for glycogen formation. To rule out this possibility, Rats 145, 146, and 147 received the glucose in three doses at hourly intervals, thus affording glucose for absorption over the entire 3 hour period. There were no appreciable differences in either liver or body glycogen between these animals which received glucose in fractional doses and those which received the entire amount of glucose in a single feeding. The lack of significant deposition of glycogen after xylose feeding can hardly be due to the slow absorption of this sugar but must in all probability be related to the difficulty of utilization of the xylose.

SUMMARY

1. The absorption of xylose from the gastrointestinal tract has been determined by the method of Cori (8) in the white rat. In confirmation of the work of earlier investigators, xylose was absorbed more slowly than glucose. The rate of absorption under the experimental conditions used was lower during the 1st hour than in subsequent hours, absorption coefficients of 29, 39, and 46 mg. per 100 gm. of rat per hour being obtained for absorption periods of 1, 2, and 3 hours duration.

2. No evidence was obtained that xylose fed under our experimental conditions was a significant source of glycogen in the white rat. A marked glycogenesis was observed in experiments in which glucose was fed in comparable amounts and under similar conditions. These studies afford no evidence of utilization of xylose in the rat.

BIBLIOGRAPHY

1. Koch, E., *Pharm. Z. Russland*, **25**, 657 (1886); *Ber. chem. Ges., Ref.*, **20**, 145 (1887).
2. Schreiber, W. T., Geib, N. V., Wingfield, B., and Acree, S. F., *Ind. and Eng. Chem.*, **22**, 497 (1930).
3. *Literary Digest*, **105**, 30 (1930).
4. Pfüger, E. F. W., *Das Glykogen und seine Beziehungen zur Zuckerkrankheit*, Bonn, 2nd edition, 230 (1905).
5. Brasch, W., *Z. Biol.*, **50**, 113 (1907-08).
6. Thomas, P., Gradinescu, A., and Imas, R., *Compt. rend. Acad.*, **188**, 664 (1929).
7. Nitzescu, I. I., and Benetato, M., *Compt. rend. Soc. biol.*, **102**, 175 (1929).
8. Cori, C. F., *J. Biol. Chem.*, **66**, 691 (1925).
9. Wilson, R. H., and Lewis, H. B., *J. Biol. Chem.*, **85**, 559 (1929-30).
10. Macleod, J. J. R., Magee, H. E., and Purves, C. B., *J. Physiol.*, **70**, 404 (1930).

PENTOSE METABOLISM

II. THE PENTOSE CONTENT OF THE TISSUES OF THE WHITE RAT AFTER THE ORAL ADMINISTRATION OF *d*-XYLOSE

BY MABEL M. MILLER AND HOWARD B. LEWIS

(From the Department of Physiological Chemistry, Medical School, University of Michigan, Ann Arbor)

(Received for publication, July 8, 1932)

In the preceding paper (1), a study has been made of the rate of absorption of xylose from the gastrointestinal tract of the young white rat by the method of Cori and of the formation of glycogen after xylose feeding. The xylose was shown to be absorbed readily, but at a rate much less rapid than was glucose. Under the experimental conditions, no formation of glycogen could be demonstrated after the oral administration of xylose. In an attempt to gain further information as to the fate of the xylose absorbed, we have been concerned with the changes in the pentose content of the tissues (liver, kidney, muscle, blood) after the feeding of xylose.

EXPERIMENTAL

The experimental animals, young white rats, were fasted for 24 hours and were then fed xylose (2 cc. of a 50 per cent solution) as already described (1) in the preceding paper. After an absorption period of 3 hours, the animals were killed by chloroform. The liver, kidneys, and muscles of the hind legs were removed with as little injury to the tissues as possible, rinsed with a very small amount of water to remove any surface blood, wiped dry with filter paper, and weighed as rapidly as possible.

Protein-free extracts for the determination of pentoses were prepared as follows: Liver or kidney tissue was minced with scissors and ground in a mortar with clean quartz sand and 8 cc. of 10 per cent trichloroacetic acid. The extract obtained was decanted through a small filter paper into a 50 cc. volumetric flask and the grinding and extraction were repeated twice with fresh trichloro-

acetic acid, a total of 25 cc. of the acid being used. The mortar was then rinsed twice with 8 cc. of water and the rinsings were transferred to the filter. The filter paper was washed with water to give a final volume of 50 cc. in the volumetric flask. The procedure with muscle tissue was similar except that the minced tissue was ground with sand and 10 cc. of water before the addition of the first portion of trichloroacetic acid. The entire liver, both kidneys, and samples of approximately 2 gm. of muscle were used for the analyses.

The blood (removed by puncture from the heart) was transferred to a 50 cc. volumetric flask containing approximately 15 cc. of water. To the hemolyzed blood, 25 cc. of the trichloroacetic acid were added slowly, the flask was shaken vigorously, the contents were diluted to the mark with water, shaken, and the precipitated proteins were removed by filtration.

Pentoses were determined in the protein-free extracts by the colorimetric method of McCance (2), modified slightly because of the presence of trichloroacetic acid in our filtrates. In routine determinations, 4 cc. of protein-free filtrate from liver were measured into a Pyrex test-tube and 2 cc. of water and 3 cc. of concentrated hydrochloric acid were added. In the standard tube were placed 2 cc. of a xylose solution containing 0.1 mg. of xylose per cc., 2 cc. of 10 per cent trichloroacetic acid (since each 4 cc. of the tissue extract contained this amount of trichloroacetic acid), 2 cc. of water, and 3 cc. of concentrated hydrochloric acid. In the case of filtrates from kidney, muscle, and blood, the pentose content was usually too low to give sufficient color in the reaction to read satisfactorily. To 4 cc. of these filtrates 2 cc. of the standard xylose solution (0.2 mg. of xylose) were added in place of the water.

The Pyrex test-tubes containing the unknowns and the standards were fitted with reflux condensers consisting of 24 inch glass tubes inserted through tin-foil covered corks. The tubes containing the condensers were placed in a boiling water bath for 2 hours. After removal from the water bath, the tubes were cooled and 4 cc. of benzene were added to each. The tubes were then corked and shaken vigorously for 3 minutes in order to extract the furfural into the benzene layer. The tubes were allowed to stand for 30 minutes to get a complete separation of the benzene layer from the water layer, 2 cc. of the benzene layer were carefully removed, and 3 cc.

of the benzidine solution were added to develop the color. If any water became mixed with the benzene, the color would not develop completely. To further protect against the presence of moisture, all test-tubes and corks used for aliquots of the benzene layer were dried in the oven and kept in a desiccator until used. In spite of these precautions, great difficulty with the fading of the colors or with the lack of complete color development was experienced. The cause was found to lie in impurities present in some of the reagents used. Therefore, all reagents were purified. The trichloroacetic acid was distilled twice, the benzidine was recrystallized twice from water-free benzene, dried at 40°, and kept in a desiccator until needed. Thiophene-free benzene was used and a satisfactory lot of glacial acetic acid was found by testing the various samples available in the laboratory. The absolute alcohol and glacial acetic acid were tested with anhydrous copper sulfate for the presence of water and were not used if any blue color developed. After all of these precautions had been taken, the color developed fully within 15 minutes and showed no evidence of fading throughout a 2 hour period.

The accuracy of the method was tested with xylose solutions of known concentration and with mixtures containing xylose and glucose. The best proportionality was obtained with solutions of such a strength that the colorimetric reading was slightly lower than that of the standard, but good proportionality was secured with solutions of widely different strength. No interference in the development of the color resulted from the presence of glucose. It was also necessary to determine whether xylose added to tissue extracts could be recovered satisfactorily since xylose was added to certain of the tissue extracts in order to obtain a color of sufficient intensity to permit accurate comparison in the colorimeter. A typical series of control experiments of this kind may be cited. The pentose content of 2 cc. of a trichloroacetic acid filtrate of liver was found to be 0.157 mg. When 0.05, 0.10, and 0.15 mg. of xylose were added to 2 cc. portions of this filtrate, the pentose determinations showed 0.204, 0.259, and 0.308 mg. respectively or a pentose content of the original filtrate of 0.154, 0.159, and 0.158 mg. respectively as compared with the value of 0.157 mg. for the filtrate to which no xylose was added. That is, the pentose content of the liver extract was essentially the same regardless of

whether xylose was added to the extract. While we realize that the addition of known amounts of xylose to the unknown and the calculation of the xylose content of the tissue extracts by difference are not desirable procedures, we believe that the results of a long series of similar check experiments justify this, particularly since in small animals such as those used here, the amount of material available for analysis is small. Moreover, the difference in the analytical results between our control and our experimental animals is clearly beyond the possible error of the method.

It is recognized that the values obtained by this procedure cannot be interpreted strictly as pentoses. Glycuronic acid gives rise to furfural on treatment with acid. However, in a trichloroacetic acid filtrate from tissues, glucoproteins should not be present and glycuronic acid should exist only in the form of free chondroitic acid or similar substances or as conjugated glycuronates. Furfural may also arise from combined pentoses of tissues. Nucleoproteins should not occur in our protein-free filtrates, but the presence of nucleotides and nucleosides appears probable. The presence of nucleotides (*e.g.* adenylic, guanylic, and inosinic acids) in blood and tissues has frequently been demonstrated. Nucleotides of different origin vary greatly in their stability. Thus muscle adenylic acid (3) has been shown to yield furfural in traces only by the pentose method of Hoffman (4), while yeast adenylic acid yields furfural abundantly (3, 4). The pyrimidine nucleotides are relatively stable and yield little furfural, while the purine nucleotides and nucleosides (from yeast) yield furfural almost quantitatively (4). We have observed a similar ease of formation of furfural from the pentose present in yeast nucleic acid and adenine mononucleotide from yeast¹ in the McCance pentose method as used by us. It is probable that some of the pentose measured in our determination may have originated from non-pentose precursors and that some may have been combined as nucleoside or nucleotide. We have felt it permissible to speak of our results in terms of "pentose" or of "xylose" since our chief interest lies in the comparison of the results obtained in control experiments and in experiments in which xylose was fed.

¹ We are indebted to Dr. H. O. Calvery for supplying us with these substances for a study of their behavior in the method for pentose determination.

DISCUSSION

The experimental results obtained with rats fasted 24 hours and with rats fed xylose 3 hours prior to death are presented in Table I. In the fasting control animals, the pentose content of the tissue extracts was higher in muscle tissue than in liver or kidney. We have been unable to find data on the pentose content of tissues which are comparable to our own. Direct comparison with the few values in the literature cannot be made since in our experiments, the analyses were made on protein-free extracts of the tissues, whereas the older determinations were made on the hydrolysis products of the entire tissue. Moreover, Bendix and Ebstein (5) dried the tissues prior to analysis by shaking with alcohol, ether, and acetic acid, a procedure which might have been expected to remove soluble pentoses either free or in combination. In general, our values for liver and kidney, as might be expected, are lower than those previously recorded for the total pentose content of tissues (5-8). In the case of muscle tissue, which contains such soluble substances as inosinic and adenylic acids, our values were higher than those obtained on washed tissue.

In the xylose feeding experiments, after an absorption period of 3 hours, the pentose contents of the liver and kidney were greatly increased (Table I). The experimental values indicate definite increases since in every case the minimal values for these tissues of the xylose-fed rats were greater than the maximal values of the control series. The range was greater in the values obtained for the kidney than for the liver. This is probably to be explained by the accumulation of the pentose in the kidney prior to excretion in the urine, since it has long been recognized that after ingestion of pentoses, an excretion of these carbohydrates occurs. In contrast to those of the liver and kidney, the pentose contents of the muscles were practically identical in the control animals and in the animals receiving xylose. Cori and Goltz (9) have shown that arabinose penetrates muscle tissue more slowly than liver, a finding in harmony with the results of these experiments.

In order to demonstrate that these changes in the pentose content of the tissues were due to the pentose fed, a control series was studied in which 2 cc. of a 50 per cent solution of glucose were fed in place of xylose. In contrast to the experiments just discussed,

the results (Table II) showed clearly that the administration of glucose did not alter the pentose content of the tissues.

TABLE I

"Pentose" Content of Liver, Muscle, and Kidney of White Rats after a 24 Hour Fast and after Oral Administration of Xylose

All results are calculated as mg. of xylose per 100 gm. of tissue.

	Rat No.*	Weight after fast	Pentose		
			Liver	Kidney	Muscle
		gm.	mg.	mg.	mg.
Fasted control rats	160	163	56	34	126
	162	163	65	50	88
	164	149	58	49	99
	166	156	56	46	125
	172	170	51	50	81
	175	167	61	66	90
	214	162	40	37	92
	219	171	56	45	81
	168	165	41	71	86
	169	178	54	42	88
	170	164	43	44	49
	171	158	48	68	88
Average.....			53	50	91
Xylose-fed rats	161	154	83	94	105
	163	167	88	107	95
	165	144	86	116	113
	167	150	105	121	133
	173	170	73	83	83
	176	175	92	128	93
	215	162	83	103	88
	218	148	99	126	89
	177	189	95	134	76
	178	177	93	120	81
	179	205	88	120	88
	180	182	81	109	62
Average.....			89	113	92

* The determinations with the first eight rats in each group were made in pairs, a fasting control experiment being carried out at the same time as a xylose feeding experiment. Thus the experiments with Rats 160 and 161, with Rats 162 and 163, etc. represent paired experiments.

It has been maintained by some workers that the administration of pentoses results in a mobilization of glucose. Blanco (10), in a study of the glucose content of the blood by the method of Somogyi, noted definite increases in the glucose of the blood 3 to 3.5 hours after oral or intravenous administration of xylose; the results after subcutaneous injection were variable. We have determined total reducing substances in trichloroacetic acid filtrates of the blood by the method of Hagedorn and Jensen and

TABLE II

"Pentose" Content of Tissues of Rats after a 24 Hour Fast and after Glucose Feeding

All results are calculated as mg. of xylose per 100 gm. of tissue.

	Rat No.*	Weight after fast gm.	Pentose			
			Liver	Kidney	Muscle	Blood
			mg.	mg.	mg.	mg.
Fasted control rats	239	137	45	56	91	11
	240	148	50	41	55	7
	242	156	50	50	71	7
	244	156	54	59	63	6
Average.....			50	51	70	8
Rats fed glu- cose	238	134	46	49	80	11
	241	157	48	47	54	Lost
	243	155	49	40	74	8
	245	154	46	57	65	7
Average.....			47	48	68	9

* The procedure of each feeding experiment was checked by a fasting control experiment carried out simultaneously. Thus the experiments with Rats 239 and 238, with Rats 240 and 241, etc. are paired experiments.

have calculated the values obtained in terms of xylose. We have also estimated the xylose content (pentose) of these blood filtrates by the McCance method already described. The figures obtained with fasting control animals and with animals killed 3 hours after xylose feeding are presented in Table III. By subtracting the values for xylose from the total reducing substances, we have obtained approximate values for total reducing substances not xylose.

The pentose (xylose) content of the blood as determined by the

TABLE III

Composition of Blood in Fasted Rats and in Rats after Feeding of Xylose

All results are expressed as mg. per 100 cc. of blood calculated as xylose.

	Rat No.	Group No.*	Total reducing substances (a)	Xylose (b)	Total reducing substances not xylose (a - b)
			mg.	mg.	mg.
Fasted control rats	220	1	125	9	116
	221	1	107	10	97
	225	2	122	7	115
	226	2	90	6	83
	227	2	115	8	106
	231	3	100	7	92
	232	3	116	7	109
	233	3	122	10	112
	234	3	140	7	132
	248	4	135	6	128
	249	4	106	8	99
	250	4	112	7	105
Average.....			116	7	108
Rats fed xylose	222	1	145	30	115
	223	1	145	30	115
	224	1	142	30	112
	228	2	187	39	148
	229	2	176	33	143
	230	2	172	40	132
	235	3	175	32	142
	236	3	147	34	113
	237	3	190	34	156
	251	4	175	31	143
	252	4	150	30	119
	253	4	155	34	121
Average.....			163	33	130

* The analyses of the blood of all rats (experimental and control) of the same group were carried out simultaneously.

method of McCance showed a considerable and remarkably uniform increase 3 hours after the xylose feeding. Corley (11) who fed *d*-xylose to rabbits in amounts ranging from 1 to 3 gm. per kilo. amounts smaller than those fed in our experiments, observed

increases in the unfermentable reducing substances of the blood, which were much more marked than those observed in the present study with rats.

The total reducing substances not xylose (Table III) showed a very slight tendency to increase after a 3 hour xylose absorption period. The results were too variable and the increases too slight in comparison with the fasting control values to be considered entirely significant. Further work more carefully controlled is necessary to determine whether these slightly increased values are significant or within the limits of error of our experimental procedure.

The experimental data obtained in this and the preceding paper are being extended. Further studies on the nutritive value and utilization of the pentoses are in progress in this laboratory.

SUMMARY

1. The method of McCance has been adapted to the determination of the pentose content of trichloroacetic acid filtrates of tissues and blood.

2. After oral administration of *d*-xylose to the white rat, the pentose contents of trichloroacetic acid extracts of liver, kidney, and blood were increased. Under the same experimental conditions no changes in the pentose values of trichloroacetic acid extracts of muscle were observed.

3. The oral administration of glucose did not influence the pentose content of the tissues studied.

4. The increase in the reducing substances of the blood, other than those which yielded furfural on treatment with hydrochloric acid (*e.g.* pentoses), after a 3 hour absorption period after xylose feeding, was so slight as to be almost within the range of variation of the values for the control animals.

BIBLIOGRAPHY

1. Miller, M. M., and Lewis, H. B., *J. Biol. Chem.*, **98**, 133 (1932).
2. McCance, R. A., *Biochem. J.*, **20**, 1111 (1926).
3. Embden, G., and Schmidt, G., *Z. physiol. Chem.*, **181**, 130 (1929).
4. Hoffman, W. S., *J. Biol. Chem.*, **73**, 15 (1927).
5. Bendix, E., and Ebstein, E., *Z. allg. Physiol.*, **2**, 1 (1902-03).
6. Grund, G., *Z. physiol. Chem.*, **35**, 111 (1902).

7. Mancini, S., *Arch. farmacol. sper.*, **5**, 309, 337 (1906); *Chem. Zentr.*, **2**, 892 (1906).
8. Berkeley, C., *Tr. Roy. Soc. Canad.*, **15**, sect. 5, 41 (1921); *J. Biol. Chem.*, **58**, 611 (1923-24).
9. Cori, C. F., and Goltz, H. L., *Proc. Soc. Exp. Biol. and Med.*, **23**, 124 (1925).
10. Blanco, J. G., *J. Biol. Chem.*, **79**, 667 (1928).
11. Corley, R. C., *J. Biol. Chem.*, **76**, 23 (1928).

WAVERLY PRESS, INC.
BALTIMORE, U. S. A.